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(54) FLUORESCENCE FILTERING SYSTEM AND METHOD FOR MOLECULAR IMAGING

FLUORESCENZFILTERSYSTEM UND VERFAHREN ZUR MOLEKULAREN ABBILDUNG

SYSTEME DE FILTRATION PAR FLUORESCENCE ET PROCEDE D'IMAGERIE MOLECULAIRE

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Description

Background

[0001] A fluorescence optical system illuminates a fluorophore-labeled target with light whose wavelength content falls within the absorption band and collects light whose wavelength content is in the emission band. An emission filter placed in front of a detector filters light that is not in the emission band. One challenge with emission filters is that unwanted photon rejection depends on the angle at which light traverses the filter. Specifically, as the angle of incidence increases, the transmission/reflection of the filter shifts to lower wavelengths. Accordingly, even if the field of view is a single point that provides an axial ray at a 0 degree angle, other rays of the same light beam will pass through the filter at non-0 degree angles and, accordingly, may experience different amounts of filtering.

[0002] This situation is addressed in Hwang et al., "The influence of improved interference filter performance for molecular imaging using frequency domain photon migration measurements," Optical Tomography and Spectroscopy of Tissue VI, SPIE vol. 5693, pp. 503-512. Hwang et al. describes an optical system in which a collimator is placed between imaging optics and an emission filter. The collimator ensures that all rays in a light beam originating from a certain point in the image field will pass through the filter at a 0 degree angle and, thus, will receive the same type of filtering. However, if a relatively large field of view is used, light beams emanating from the edge of the field, while still collimated, will pass through the filter at an angle. This results in different amounts of excitation leakage across the field.

[0003] There is a need, therefore, for a fluorescence filtering method and system that will overcome this problem.

[0004] In US 5 206 699 A diamonds are detected by passing the emitted radiation through a narrow band pass filter and sensing the Raman radiation with a photo-multiplier tube. In front of the photo-multiplier tube a filter is positioned and in front of the filter a lens.

[0005] In US 5 943 129 A a system is disclosed comprising a light source, a filter that transmits light of a selected spectral range. The light passing the filter is directed onto a sample that is capable of responsively fluorescing and emitting light when so illuminated. A light detector is provided with in front of the light detector a lens and in front of the lens a filter.

[0006] US2004/125370 A1 discloses a device for optical examination of biological material arranged to illuminate a specific pixel by focusing excitation light and analyzing the light emitted from this pixel using a detector. The full field of view of the biological material is screened by moving the table onto which is applied the biological material.

[0007] US 6,345,115 B1 discloses an imaging system where the field of view is illuminated using a ring light

system in order to maximize the light transfer from the lens to the light detector. To that purpose, an object telecentric space is created.

Summary

[0008] The present invention is defined by the following claims.

[0009] By way of introduction, it is disclosed a method for fluorescence filtering and a fluorescence filtering system for molecular imaging comprising a source subsystem comprising a light source and a set of excitation filters designed to pass wavelengths of light in an absorption band of a fluorescent material. A detector system is also disclosed comprising a light detector, imaging optics, a first set of filters designed to pass wavelengths of light in an emission band of the fluorescent material, and an aperture located at a front focal plane of the imaging optics. A telecentric space is created between the light detector and the imaging optics, such that axial rays from a plurality of field points emerge from the imaging optics parallel to each other and perpendicular to the first set of filters. Other preferred embodiments are provided, and each of the preferred embodiments described herein can be used alone or in combination with one another within the scope of the appended claims.

[0010] The preferred embodiments will now be described with reference to the attached drawings.

Brief Description of the Drawings

[0011]

Figures 1A and 1B are graphs showing wavelength shifting of a band-pass filter due to incident angular variation.

Figure 2 is an illustration of an optical arrangement in which an emission filter is placed in front of imaging optics.

Figure 3 is an illustration of an optical arrangement in which an emission filter is placed between the imaging optics and a detector.

Figure 4 is an illustration of an optical arrangement using a collimator.

Figure 5 is an illustration of a detector system of a preferred embodiment.

Figure 6 is an illustration of a detector system with a filter wheel of a preferred embodiment.

Figure 7 is an illustration of a fluorescence filtering system of a preferred embodiment.

Figure 8 is an illustration of a fluorescence filtering

system of another preferred embodiment.

Figure 9 is a graph of transmission curves for excitation and emission filters of a preferred embodiment.

Figure 10 is a graph showing the same data as in Figure 9 but in log scale.

Figure 11 is a graph showing reduction in residual leakage using the filtering architecture shown in Figure 7.

Figure 12 is a graph showing a horizontal cross-section from a fluorescence image obtained with a prototype system of a preferred embodiment with one band-pass filter placed in front of the lens at $T = 5$ s.

Figure 13 is a graph showing a horizontal cross-section from a fluorescence image obtained with a prototype system of a preferred embodiment with one band-pass filter placed behind the lens at $T = 5$ s.

Figure 14 is a graph showing a horizontal cross-section from a fluorescence image obtained with a prototype system of a preferred embodiment at $T = 5$ s.

Figure 15 is a graph showing a horizontal cross-section from a fluorescence image obtained with a prototype system of a preferred embodiment at $T = 120$ s.

Detailed Description of the Presently Preferred Embodiments

[0012] Fluorescence detection is a tool for molecular imaging. It enables researchers to detect particular components of complex bio-molecular assemblies, such as in live cells. Fluorescence is a photo-physical process that involves the interaction of light with certain molecules called fluorophores or fluorescent dyes. It consists of the absorption of light energy at the appropriate wavelength by such molecules and the subsequent emission of other light photons at longer wavelengths. The wavelength ranges that a fluorophore molecule can absorb and emit at are called absorption and emission bands, respectively.

[0013] A fluorescence optical system illuminates a fluorophore-labeled target with light whose wavelength content falls within the absorption band and collects light whose wavelength content is in the emission band. The source(s) and optics that generate the illumination part of the system are called the "excitation optics," and the optics used to collect the fluorescence emission are called the "emission optics." Since it is rarely possible to find a light source that has a spectral content (i.e., wavelength range) that exactly matches every fluorophore absorption band, special optical filters (usually band-pass

filters) are used along with the light sources to limit the range of illuminating wavelengths to that of the absorption band and not the emission band. At the same time, other filters are used in the emission path to allow light with wavelengths in the emission band only to reach the detector.

[0014] The task of a fluorescence optical system design is to make sure that photons with wavelengths in the absorption band only reach the target, and photons with wavelengths in the emission band only reach the detector. If not, photons from the light source will wrongly be considered as fluorescence, and, therefore, a wrong measure of the amount of fluorophore dye results. This can be a tough task if the amount of emitted fluorescence is much less than the amount of excitation light scattered by the target surface (i.e., not absorbed). This is usually the case for in-vivo imaging, such as in small animal imaging, since there are a number of challenges to achieving good signal-to-noise performance when imaging fluorescence targets deep inside small animals.

[0015] One challenge is that the amount of excitation light that reaches the inside of an animal is usually quite low because of the significant absorption and scattering caused by the various body parts (skin, muscle, fat, bone, etc.). For example, the transmission through "shaved skin + fat layer + whole rib cage + abdominal wall" is in the order of 10^{-6} and varies with the thickness and composition of each of those parts. The emitted fluorescence will have to traverse a comparable tissue path back up towards the detection system. Thus, the level of fluorescence is $\ll 10^{-12}$ times that of the excitation signal. So, for example, if a flux density of 1 mW/cm^2 impinges upon the outside of a mouse or other small animal, only a sub-nano Watt optical signal actually reaches dye-labeled cells inside the abdomen, and, in turn, only sub-femto Watt of fluorescence signal reaches the detector. The low amount of emitted fluorescence is further reduced by absorption and scattering as it makes its way out towards the detector. This means that the scattering from the excitation light that occurs at the outer parts of the animal can cause much higher levels than the fluorescence signal itself. At the same time, existing optical filter technology (e.g., thin-film emission filters, such as multi-cavity designs) can, at best, provide rejection of unwanted photons only in the order of OD6 (10^{-6}). So, standard fluorescence methods would allow through high non-fluorescent background levels and, in turn, result in low Signal-to-Background (SBR) and Signal-to-Noise (SNR) ratios.

[0016] Another challenge is that unwanted photon rejection also depends on the angle at which light traverses the filter, as the spectral properties of optical thin film filters vary with the angle of incidence of light. Specifically, as the angle of incidence increases, the transmission/reflection of the filter shifts to lower wavelengths ("blue shift"). This shift can be described by

$$\lambda(\theta) = \lambda_o \sqrt{1 - (\sin\theta/\bar{n})^2}$$

where θ is the angle deviation from the normal to the filter, and $\bar{n}$ is the effective index of refraction of the thin-film. The value of $\bar{n}$ is typically in the range of 1.5 to 2.5 and varies with polarization.

[0017] Figures 1A and 1B are graphs (transmission and transmission (dB), respectively) showing wavelength shifting of a band-pass filter due to a varying angle of incidence (0, 10, and 20 degrees). As shown in these graphs, as the angle of collected light increases relative to the normal to the filter, the effective transmission band shifts to lower wavelengths, and the amounts of transmitted fluorescence and background signals change accordingly. Light from the target spans a significant range of field angles when a relatively large field of view is imaged, such as in the case of small animal imaging. Therefore, in small animal imaging where a relatively large field of view is imaged, the resulting emission filtering (i.e., transmitted SBR) is non-constant across the image. Accordingly, it is desired to use special spectral filtering solutions in order to improve the rejection of non-fluorescence light across the whole field of view (i.e., where light is collected at different angles).

[0018] Many current area fluorescence imaging techniques use the same excitation and emission filters designed for microscopy and scanning systems and use arrangements where the emission filter 5 is placed in front of the imaging optics 10 (as in Figure 2) or behind it (as in Figure 3, where the emission filter 5 is between the imaging optics 10 and the detector 15 (here, a CCD)). (The horizontal lines from which the emission is originating in these and other figures herein represent a target, such as mouse or other small animal.) These filters are typically multi-cavity interference filters optimized for maximum rejection in the excitation band and maximum transmission in the emission band. As discussed earlier, the spectral properties of such filters vary with the angle of incidence of light. Because, in Figure 2, the axial ray 20 (i.e., the "chief" or center ray of a light beam) of light beam 25 is at a 0 degree angle to the filter 5, while the axial ray 30 of light beam 35 is at about a 45 degree angle to the filter 5, the filter 5 will provide different photon rejection characteristics of the axial rays 20, 30. This is also true in the arrangement in Figure 3. In Figure 3, the filter 5 is behind the imaging optics 10. Because the pupil plane (i.e., the plane at which axial rays of all light beams cross) is in the center of the imaging optics 10, the axial ray passes through the imaging optics 10 without changing direction. Accordingly, the filter 5 in Figure 3, like the filter 5 in Figure 2, will provide different photon rejection characteristics of the axial rays 20, 30. Accordingly, in both arrangements, the angular spectral dependence of the filter 5 results in a significant amount of excitation leakage that both limits the achievable SBR and is non-constant across the image.

[0019] It should be noted that, even in the instance where the axial ray 20 passes through the filter 5 at a 0 degree angle, other rays of the light beam 25 pass through the filter 5 at a non-0 degree angle. Accordingly, even if the field of view is a single point that provides an axial ray at a 0 degree angle, other rays of the same light beam will pass through the filter 5 at non-0 degree angles and, accordingly, may experience different amounts of filtering by the filter 5 due to the angular spectral dependence problem.

[0020] This situation is addressed in Hwang et al., "The influence of improved interference filter performance for molecular imaging using frequency domain photon migration measurements," Optical Tomography and Spectroscopy of Tissue VI, SPIE vol. 5693, pp. 503-512. Figure 4 is an illustration of the arrangement disclosed in Hwang et al. As shown in Figure 4, a collimator 40 is placed between imaging optics 45 and band-pass and holographic filters 50, 55. (Hwang suggests the use of a holographic notch filter 55 to enhance the rejection capability of the band-pass filter 50.) A lens 60 focuses the light beams passing through the filters 50, 55 onto a CCD detector 65. The collimator 40 causes the rays of each of the light beams to exit the collimator 40 parallel to each other. As a result, unlike the situation noted above, if the field of view is a single point that provides an axial ray 70 through the filters 50, 55 at a 0 degree angle, other rays of the same light beam 75 will also pass through the filters 50, 55 at a 0 degree angle because of the effect of the collimator 40. However, as shown in Figure 4, if a relatively large field of view is used, a light beam 80 emanating from the edge of the field, while still collimated, traverses the filters 50, 55 at an angle. This is because the pupil plane is in the center of the imaging optics 45, and the axial ray 85 of light beam 80 passes through the imaging optics 45 without changing direction. Accordingly, light from different field points enter the filters 50, 55 at different angles and, therefore, results in different amounts of excitation leakage across the field.

[0021] Figure 5 is an illustration of a detector system 100 of a preferred embodiment that minimizes field dependence and maximizes the Signal to Background Ratio (SBR) performance of spectral filtering. The detector system 100 comprises a light detector 105 (such as a CCD), imaging optics 110 with an equivalent focal length F, a set of filters 115 positioned between the light detector 105 and the imaging optics 110, and an aperture 120 located at a front focal plane of the imaging optics 110. As used herein, the term "imaging optics" refers to one or more optical elements whose function collectively is to project a scene onto a detector (e.g., a sensor array) such as a CCD camera. Imaging optics can comprise a single lens if its placement allows it to project the picture of a given scene onto the detector. Imaging optics can also comprise two or more lenses together in such a way that they all work together to produce the same function (i.e., project the image of a scene onto a detector). The term "imaging optics" can be used interchangeably with

the terms "imaging lens" and "imaging lens assembly." Further, imaging optics can include components other than lenses (e.g., mirrors). As also used herein, a "set" can include one or more than one member. Accordingly, a set of filters, for example, can contain a single filter or a plurality of filters. In this way, one can stack one or more filters to achieve the desired background rejection.

[0022] By locating the aperture 120 in front of the imaging optics 110, the pupil plane (i.e., the plane at which axial rays of all light beams cross) is not in the center of the imaging optics 110, and axial rays that hit the imaging optics 110 at non-0 degree angles will change direction when exiting the imaging optics 110. Further, because the pupil aperture 120 located at a front focal plane of the imaging optics 110, the pupil plane is in the front focal plane of the imaging optics 110, and a telecentric space is created between the imaging optics 110 and the light detector 105. This will cause the axial rays from a plurality of field points (i.e., locations in the imaged target) to emerge from the imaging optics 110 parallel to each other and perpendicular (i.e., at a 0-degree angle) to the set of filters 115. (A telecentric approach also eliminates otherwise unavoidable ghost images when the set of filters 115 comprises more than one filter.) As a result, each of the axial rays will receive the same filtering from the set of filters 115. While the non-axial rays of each light beam will hit the set of filters 115 at non-0 degree angles and, hence, be subject to varying filtering effects due to the angular dependence problem, such rays from each light beam will see the same effect. In other words, in the telecentric space, all the field points (light emanating from different parts of the image) traverse the set of filters 115 in the same manner, centered around the zero-degree angle. This minimizes the angular variation across the field and, thus, the resulting spectral filtering variation. Accordingly, unlike with the optical arrangement in Figure 4, light from different field points entering the set of filters 115 at different angles will result in substantially the same amount of excitation leakage across the field.

[0023] Figure 6 is an illustration of a detector system 200 of another preferred embodiment. This system 200 is similar to the system 100 in Figure 5, and common components are labeled the same. However, the system 200 in Figure 6 has an additional set of filters 210 in front of the imaging optics 110. Preferably, the set of filters 210 comprises one or more dichroic filters. This system 200 takes advantage of the fact that rays that traverse a filter placed in front of imaging optics at large angles will traverse a filter placed behind the imaging optics at smaller angles and vice versa. This has the effect of balancing out any residual leakage and, thus, flattening the field. Therefore, by placing the additional set of filters 210 in front of the imaging optics 110, the angular effect from the first set of filters 115 is balanced out more evenly across the field. Although not necessary, the additional set of filters 210 in this embodiment is located on a filter wheel 230 comprising at least one additional set of filters (not shown). Similarly, the set of filters 115 can be placed

in a filter wheel 240 comprising at least one additional set of filters (not shown). This allows different "colors" of filters to image different labels.

[0024] Turning again to the drawings, Figure 7 is an illustration of a fluorescence filtering system 300 of another preferred embodiment. This system 300 comprises a source subsystem 310 comprising two light sources 320, 330, each with a set of filters 340, 350 designed to pass wavelengths of light in an absorption band of a fluorescent material. (As discussed above, a filter may leak wavelengths of light in other bands.) The system 300 also comprises a detector subsystem 360, identical to the detector system 200 in Figure 6 (components are labeled the same). Preferably, rejection performance of the set of excitation filters 340, 350 in the excitation paths matches the rejection performance of the set of emission filters 115. Since the detector 105 responds to all the photons that pass through the excitation as well the emission bands, the rejection by both the set of excitation and emission filters 115, 340, 350 is preferably matched so that leakage from the set of excitation filters 340, 350 in the emission band will have the same effect as a comparable leakage from the set of emission filters 115 in the excitation band. It should be noted that, while Figure 7 shows two light sources 320, 330, three or more light sources can be used. Also, the number of light sources does not have to match the number of sets of filters. For example, one can use one light source with one filter set and then split the output to act like separate sources. Alternatively, one can split the output to more than one port and put filter sets in front of each port.

[0025] Figure 8 is an illustration of an alternate system 400, in which a single source 410 is used with a dichroic splitter 420. The dichroic splitter 420 is positioned such that light from the light source 410 illuminates a target and light emitted from the target reaches the detector 430. The dichroic splitter 420 also has filtering properties like the set of filters 210 in Figures 6 and 7. However, the advantage of using the set of filters 210 in Figures 6 and 7 is that they prevents any possible specular reflections from getting into the collection optics.

[0026] In one presently preferred embodiment of the system 300 shown in Figure 7, the detector is a Hamamatsu ORCA_AG detector, the imaging optics 110 is a Canon 50mm/ F2.0 lens, the set of emission filters 115 are Omega 822DF20 filters, and the second set of filters 210 is a Semrock 800LP filter, operating at a nominal zero-degree angle of incidence. The excitation sources preferably consist of two fiber-coupled, symmetrically-positioned identical laser diode sources (782nm) as the light sources 320, 330 and a set of two excitation filters 340, 350 in front of each laser 320, 330. Both excitation and emission filters have about OD6 rejection each.

[0027] Turning again to the drawings, Figure 9 is a graph showing transmission curves for the excitation and emission filters. Figure 10 shows the same data in log scale so that the rejection level can be better evaluated. Tests were conducted to confirm that rejection with a

configuration of (2, 2) excitation and emission filter sets is better than (1, 1), (1, 2), and (2, 1) configurations. Of course, if further rejection is needed, one can use (3, 3), (4, 4), etc. Figure 11 is a graph showing reduction in residual leakage from the filtering architecture shown in Figure 7. A comparison between Figures 10 and 11 show the theoretical level of reduction in background leakage that can be achieved by doubling the rejection capability of both the excitation and emission filters.

[0028] Figures 12-15 show horizontal cross-sections from images obtained with the prototype system described above. The target is a nitro-cellulose membrane with 5 IRDye®800 labeled fluorescent spots. The membrane produces a significant amount of scattering from the excitation laser and is thus used to obtain a measure of the rejection capability of the filters and the flatness of the residual background. The cross-section is arbitrarily chosen to pass through a fluorescent spot located near the center of the image. Such fluorescent spot is used to measure the fluorescence transmission efficiency. This way, a measure of Signal-to-Background (SBR) can easily be obtained. In each figure, the graph is displayed in log-scale in order to enhance the levels of the background.

[0029] In Figures 12 and 13, only one emission filter was placed in front and in the back of the lens, respectively. This is similar to what is done in most prior small animal imaging solutions. It also shows how the non-flatness of the background in both cases complements each other, and, therefore, by placing filters on both sides of the lens, a more balanced rejection is obtained. Figures 14 and 15 show the image with filters configured according to the preferred embodiment of Figure 7. In Figure 15, the exposure time is increased to 120s in order to enhance the detection of any residual background leakage. As is clear from the image, even though the fluorescent signal is much higher than saturation, the leakage is still flat and non-significant. The SBR improvement in this case is estimated to be ~ 30x.

[0030] There are several alternatives that can be used with these disclosures. For example, while the preferred invention illustrated above with respect to an application for fluorescence filtering for molecular imaging, these arrangements can be used in any suitable application. Accordingly, the filters do not have to be designed to pass wavelengths of light in absorption and emission bands of fluorescent materials. Also, while these disclosures were illustrated in terms of imaging a small animal, such as a mouse, they can be used to image other targets. Additionally, any suitable light source, detector, filter, imaging optics, and aperture can be used. Further, any of the embodiments disclosed herein can be used by itself or in combination with any of the other embodiments disclosed herein. Finally, each of the excitation filter sets can pass wavelengths in more than one excitation band, and emission filter sets can pass wavelengths in more than one emission band. Also, any of the sets of filters disclosed herein can be placed on a filter wheel.

[0031] The foregoing detailed description is to be understood as an illustration of selected forms that the invention can take and not as a definition of the invention. The following claims, including all equivalents, define the scope of this invention.

Claims

1. A detector system comprising:

- a light detector (105);
- imaging optics (110);
- a first set of filters (115), designed to pass wavelength of light in an emission band of a fluorescent material, positioned between the light detector and the imaging optics;

characterized in that

- an aperture (120) is located at a front focal plane (F) of the imaging optics (110), wherein a telecentric space is created between the light detector (105) and the imaging optics (110), such that non-parallel axial rays from a plurality of field points emerge from the imaging optics parallel to each other and perpendicular to the first set of filters.

2. The detector system of claim 1, wherein a second set of filters (210) is located on the opposite side of the imaging optics compared to the first set of filters (115).

3. The detector system of Claim 2, wherein the second set of filters (210) comprises a dichroic filter.

4. The detector system of Claim 2 or 3, wherein the second set of filters (210) is located on a filter wheel (230) comprising at least one additional set of filters.

5. A fluorescence filtering system comprising:
a source subsystem (310) comprising:

- a light source (320, 330); and
- a set of excitation filters (340, 350) designed to pass wavelengths of light in an absorption band of a fluorescent material; and
- a detector system (360) according to claim 1.

6. The fluorescence filtering system of Claim 5, wherein rejection performance of the set of excitation filters (340, 350) matches rejection performance of the first set of filters (115).

7. The fluorescence filtering system of Claim 5 or 6 further comprising a dichroic splitter (420) between the light source (410) and the detector (430), the dichroic

splitter positioned such that light from the light source illuminates a target and light emitted from the target reaches the detector.

8. A method for fluorescence filtering, the method comprising:

(a) illuminating a target comprising a fluorescent material with light in an absorption band of the fluorescent material, wherein, in response to absorbing the light in the absorption band, the fluorescent material emits light in an emission band of the fluorescent material;

(b) causing non-parallel axial rays of light beams from a plurality of field points in the target, after passing through an aperture located at a front focal plane of an imaging optics, to emerge from the imaging optics (110) parallel to each other and perpendicular to a first set of filters designed (115) to pass wavelengths of light in the emission band and (c) detecting light passed through the first set of filters (115).

9. The method of Claim 8 further comprising, before (a), passing light from a light source (320, 330) through a set of excitation filters (340, 350) designed to pass light in the absorption band.

10. The method of Claim 9, wherein rejection performance of the first set of filters (115) designed to pass wavelengths of light in the emission band matches rejection performance of the set of excitation filters (340, 350) designed to pass wavelengths of light in the absorption band.

11. The method of Claim 10, further comprising, between (a) and (b), passing the light emitted from the fluorescent material through a second set of filters (210) designed to pass wavelength of light in the emission band.

12. The method of claim 11, wherein the second set of filters (210) comprises a dichroic filter.

13. The method of Claim 11 or 12, wherein the second set of filters (210) are located on a filter wheel (230) comprising at least one additional set of filters.

14. The method according to any of Claims 11 to 13, wherein (a) is performed by a single light source.

15. The method according to any of Claims 11 to 13, wherein (a) is performed by two or more light sources.

Patentansprüche

1. Detektorsystem, umfassend:

- einen Lichtdetektor (105);
- eine Abbildungsoptik (100);
- einen ersten Filtersatz (115), der dazu ausgelegt ist, eine Licht-Wellenlänge in ein Emissionsband eines fluoreszierenden Materials übergehen zu lassen, das zwischen dem Lichtdetektor und der Abbildungsoptik angeordnet ist;

dadurch gekennzeichnet, dass

- eine Öffnung (120) an einer vorderen fokalen Fläche (F) der Abbildungsoptik (110) angeordnet ist, wobei ein telezentrischer Raum zwischen dem Lichtdetektor (105) und der Abbildungsoptik (110) derart hergestellt ist, dass nicht parallele axiale Strahlen von einer Vielzahl von Feldpunkten aus der Abbildungsoptik hervortreten, die zueinander parallel und zu dem ersten Filtersatz lotrecht sind.

2. Detektorsystem gemäß Anspruch 1, wobei ein zweiter Filtersatz (210) auf der entgegengesetzten Seite der Abbildungsoptik verglichen mit dem ersten Filtersatz (115) angeordnet ist.

3. Detektorsystem gemäß Anspruch 2, wobei der zweite Filtersatz (210) einen dichroitischen Filter umfasst.

4. Detektorsystem gemäß Anspruch 2 oder 3, wobei der zweite Filtersatz (210) auf einem Filterrad (230) angeordnet ist, umfassend wenigstens einen zusätzlichen Filtersatz.

5. Fluoreszenz-Filtersystem, umfassend:
ein Quellen-Teilsystem (310), umfassend:

- eine Lichtquelle (320; 330) und
- einen Satz Erregungsfilter (340, 350), die dazu bestimmt sind, Licht-Wellenlängen in einem Absorptionsband eines fluoreszierenden Materials hindurchtreten zu lassen; und
- ein Detektorsystem (360) gemäß Anspruch 1.

6. Fluoreszenz-Filtersystem gemäß Anspruch 5, wobei die Abweisungsleistung des Satzes Erregungsfilter (340, 350) der Leistung des ersten Filtersatzes (115) entspricht.

7. Fluoreszenz-Filtersystem gemäß Anspruch 5 oder 6, weiterhin umfassend einen dichroitischen Spalter (420) zwischen der Lichtquelle (410) und dem Detektor (430), der dichroitische Spalter ist derart angeordnet, dass das Licht von der Lichtquelle ein Ziel

beleuchtet und von dem Ziel ausgegebenes Licht den Detektor erreicht.

8. Verfahren zum Fluoreszenz-Filtern, wobei das Verfahren umfasst:

a) Beleuchten eines Ziels, umfassend ein fluoreszierendes Material mit Licht in einem Absorptionsband des fluoreszierenden Materials, wobei als Antwort auf das Absorbieren von Licht in dem Absorptionsband das fluoreszierende Material Licht in einem Emissionsband des fluoreszierenden Materials ausgibt;

b) Hervorrufen von nicht parallelen axialen Strahlen von Lichtbündeln aus einer Vielzahl von Feldpunkten im Ziel nach dem Hindurchtreten durch eine Öffnung, die an einer vorderen fokalen Ebene einer Abbildungsoptik angeordnet ist, um aus der Abbildungsoptik (110) parallel zueinander und lotrecht zu einem ersten Filtersatz hervorzutreten, der dazu bestimmt ist (115), Licht-Wellenlängen in dem Emissionsband hindurchtreten zu lassen und (c) Licht zu detektieren, das durch den ersten Filtersatz (115) hindurchgetreten ist.

9. Verfahren gemäß Anspruch 8, weiterhin umfassend vor (a) das Hindurchtreten von Licht von einer Lichtquelle (320, 330) durch einen Satz Erregungsfilter (340, 350), der dazu bestimmt ist, Licht in dem Absorptionsband hindurchtreten zu lassen.

10. Verfahren gemäß Anspruch 9, wobei die Abweisungsleistung des ersten Filtersatzes (115) der dazu bestimmt ist, Licht-Wellenlängen in dem Ausgabeband hindurchtreten zu lassen, der Abweisungsleistung des Satzes Erregungsfilter (340, 350) entspricht, die dazu bestimmt sind, Licht-Wellenlängen in dem Absorptionsband hindurchtreten zu lassen.

11. Verfahren gemäß Anspruch 10, zwischen (a) und (b) weiterhin umfassend das Hindurchtreten-Lassen des von dem fluoreszierenden Material ausgegebenen Lichts durch einen zweiten Filtersatz (210), der dazu bestimmt ist, Licht-Wellenlängen in dem Emissionsband hindurchtreten zu lassen.

12. Verfahren gemäß Anspruch 11, wobei der zweite Filtersatz (210) einen dichroitischen Filter umfasst.

13. Verfahren gemäß Anspruch 11 oder 12, wobei der zweite Filtersatz (210) auf einem Filterrad (230) angeordnet ist, das wenigstens einen zusätzlichen Filtersatz umfasst.

14. Verfahren gemäß irgendeinem der Ansprüche 11 bis 13, wobei (a) durch eine einzige Lichtquelle durchgeführt wird.

15. Verfahren gemäß irgendeinem der Ansprüche 11 bis 13, wobei (a) durch zwei oder mehr Lichtquelle(n) durchgeführt wird.

Revendications

1. Système de détection comprenant :

- un détecteur de lumière (105) ;
- une optique d'imagerie (110) ;
- un premier ensemble de filtres (115), conçu pour laisser passer la longueur d'onde de la lumière dans une bande d'émission d'un matériau fluorescent, positionné entre le détecteur de lumière et l'optique d'imagerie ;

caractérisé en ce que

- une ouverture (120) est située au niveau d'un plan focal avant (F) de l'optique d'imagerie (110), dans laquelle un espace télécentrique est créé entre le détecteur de lumière (105) et l'optique d'imagerie (110), de sorte que des rayons axiaux non parallèles provenant d'une pluralité de points de champ sortent de l'optique d'imagerie parallèlement les uns aux autres et perpendiculairement au premier ensemble de filtres.

2. Système de détection selon la revendication 1, dans lequel un deuxième ensemble de filtres (210) est situé sur le côté opposé de l'optique d'imagerie par rapport au premier ensemble de filtres (115).

3. Système de détection selon la revendication 2, dans lequel le deuxième ensemble de filtres (210) comprend un filtre dichroïque.

4. Système de détection selon l'une des revendications 2 et 3, dans lequel le deuxième ensemble de filtres (210) est situé sur une roue à filtres (230) comprenant au moins un ensemble supplémentaire de filtres.

5. Un système de filtrage de fluorescence comprenant :
- un sous-système de source (310) comprenant :

- une source de lumière (320, 330) ; et
- un ensemble de filtres d'excitation (340, 350) conçus pour laisser passer les longueurs d'onde de la lumière dans une bande d'absorption d'un matériau fluorescent ; et
- un système de détection (360) selon la revendication 1.

6. Système de filtrage de fluorescence selon la reven-

- dication 5, dans lequel la performance de rejet de l'ensemble de filtres d'excitation (340, 350) correspond à la performance de rejet du premier ensemble de filtres (115).
7. Système de filtrage de fluorescence selon l'une des revendications 5 et 6, comprenant en outre un séparateur dichroïque (420) entre la source de lumière (410) et le détecteur (430), le séparateur dichroïque étant positionné de telle sorte que la lumière provenant de la source de lumière éclaire une cible et que la lumière émise par la cible atteigne le détecteur.
8. Méthode de filtrage de la fluorescence, le procédé comprenant :
- (a) illuminer une cible comprenant un matériau fluorescent avec de la lumière dans une bande d'absorption du matériau fluorescent, dans lequel, en réponse à l'absorption de la lumière dans la bande d'absorption, le matériau fluorescent émet de la lumière dans une bande d'émission du matériau fluorescent ;
- (b) faire en sorte que des rayons axiaux non parallèles de faisceaux lumineux provenant d'une pluralité de points de champ dans la cible, après être passés à travers une ouverture située au niveau d'un plan focal avant d'une optique d'imagerie, émergent de l'optique d'imagerie (110) parallèlement les uns aux autres et perpendiculairement à un premier ensemble de filtres conçus (115) pour laisser passer les longueurs d'onde de la lumière dans la bande d'émission ; et(c) détecter la lumière passée à travers le premier ensemble de filtres (115).
9. Méthode, selon la revendication 8, comprenant en outre, avant (a), de faire passer la lumière provenant d'une source de lumière (320, 330) à travers un ensemble de filtres d'excitation (340, 350) conçus pour laisser passer la lumière dans la bande d'absorption.
10. Méthode, selon la revendication 9, dans laquelle la performance de rejet du premier ensemble de filtres (115) conçu pour laisser passer des longueurs d'onde de lumière dans la bande d'émission correspond à la performance de rejet de l'ensemble de filtres d'excitation (340, 350) conçu pour laisser passer des longueurs d'onde de lumière dans la bande d'absorption.
11. Méthode, selon la revendication 10, comprenant en outre, entre (a) et (b), de faire passer la lumière émise par le matériau fluorescent à travers un second jeu de filtres (210) conçu pour laisser passer la longueur d'onde de la lumière dans la bande d'émission.
12. Méthode, selon la revendication 11, dans laquelle le deuxième ensemble de filtres (210) comprend un filtre dichroïque.
13. Méthode, selon la revendication 11 ou 12, dans laquelle le deuxième ensemble de filtres (210) est situé sur une roue à filtres (230) comprenant au moins un ensemble supplémentaire de filtres.
14. Méthode selon l'une quelconque des revendications 11 à 13, dans laquelle (a) est réalisé par une seule source de lumière.
15. Méthode selon l'une quelconque des revendications 11 à 13, dans laquelle (a) est réalisé par deux ou plusieurs sources de lumière.

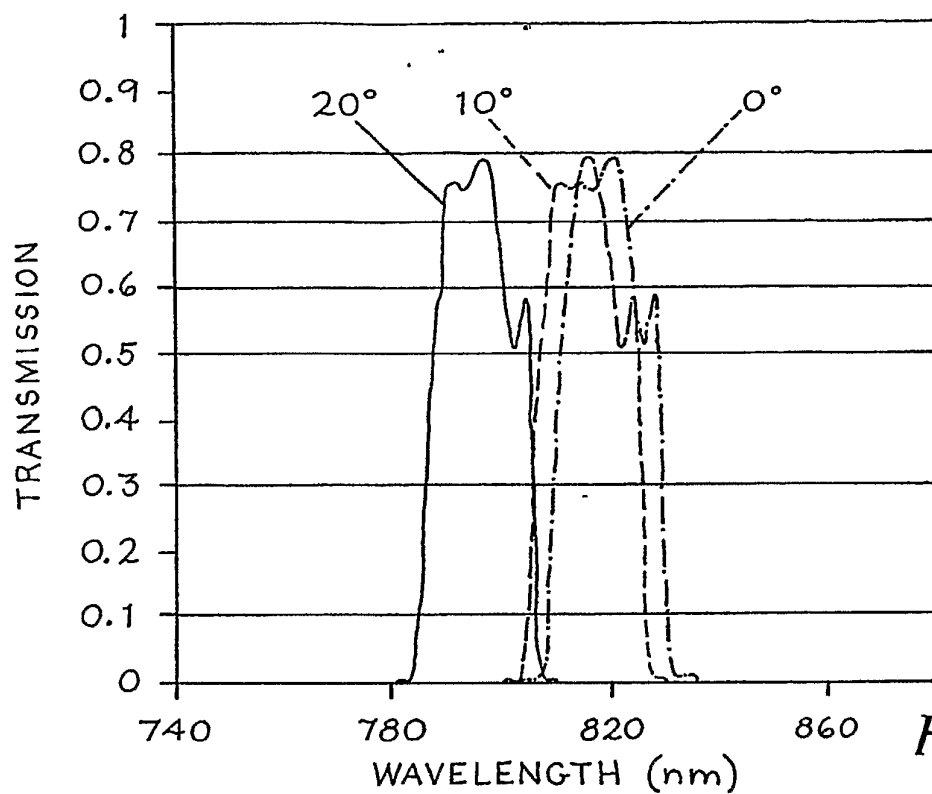


Fig. 1A

--- 0 DEGREES -- 10 DEGREES — 20 DEGREES

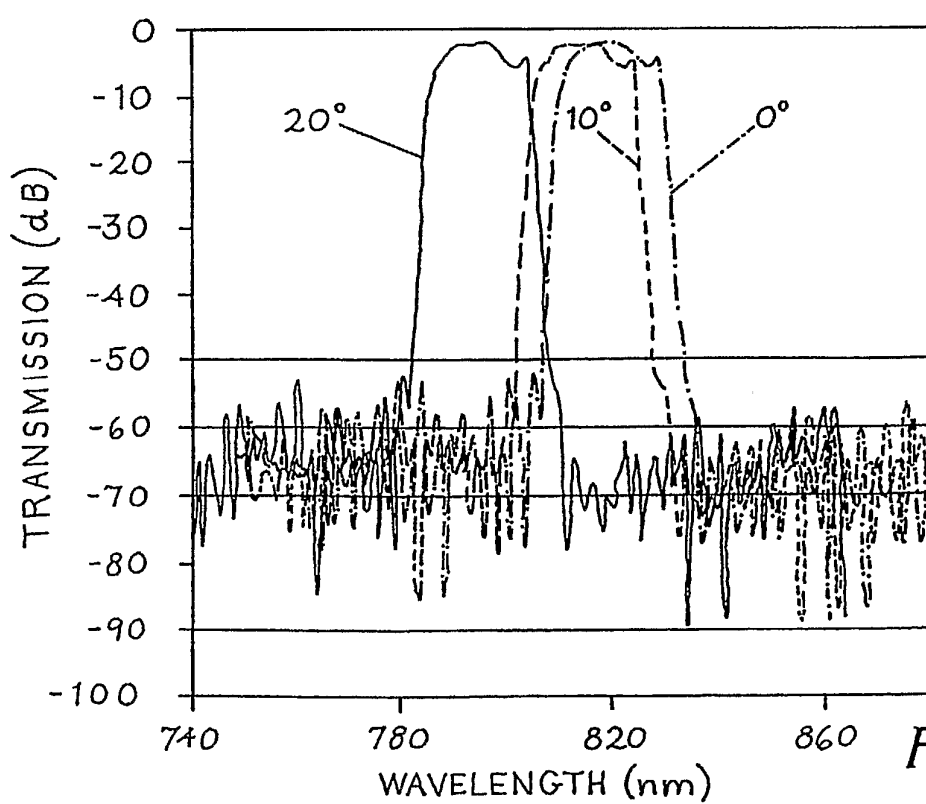
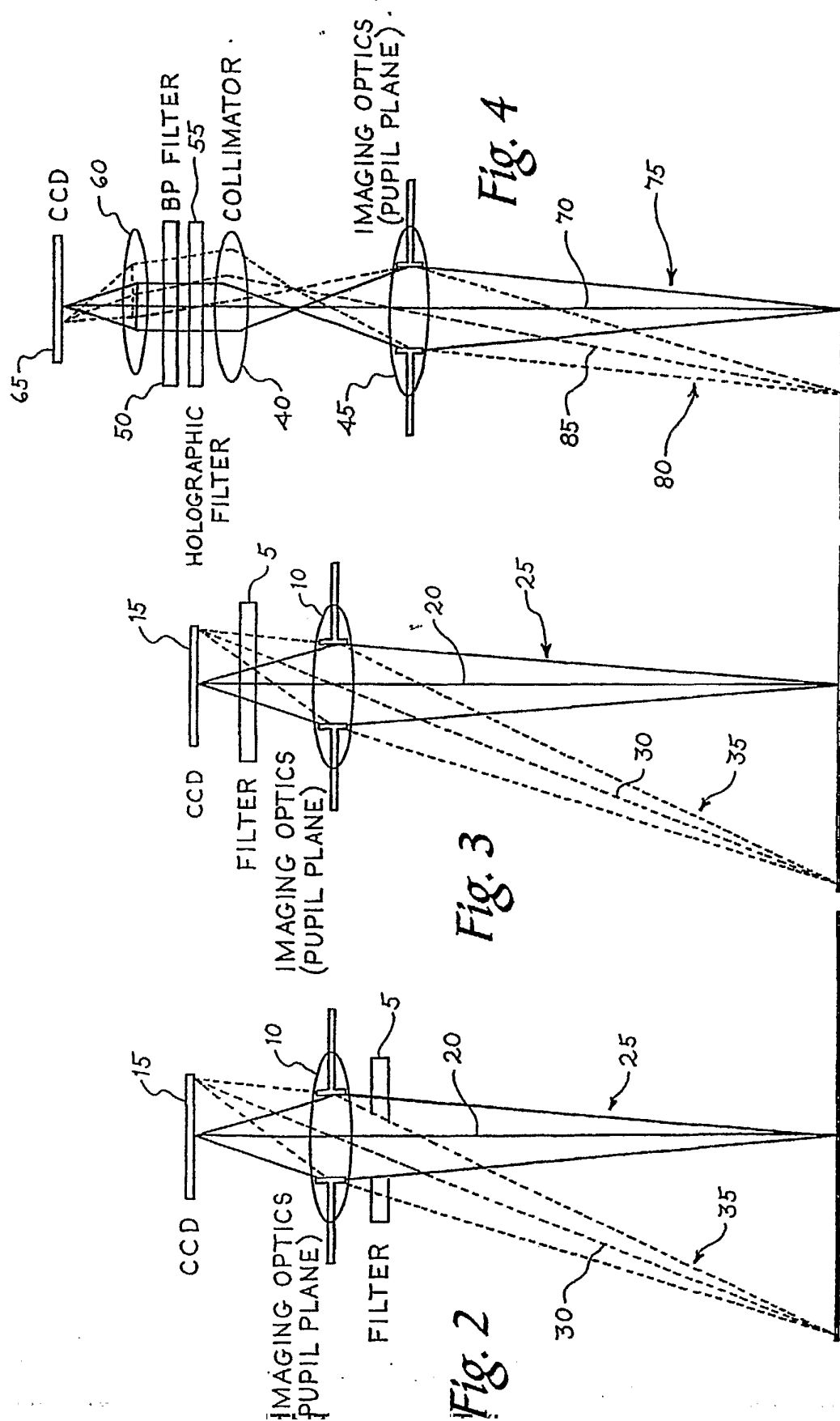
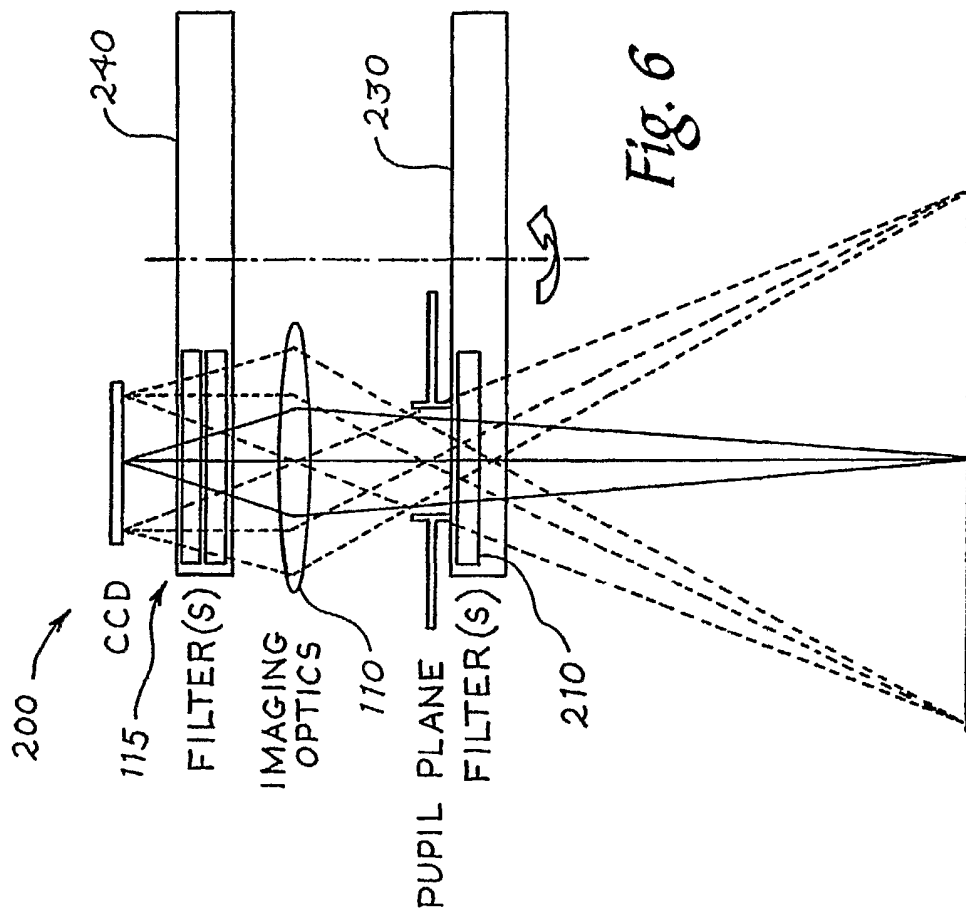
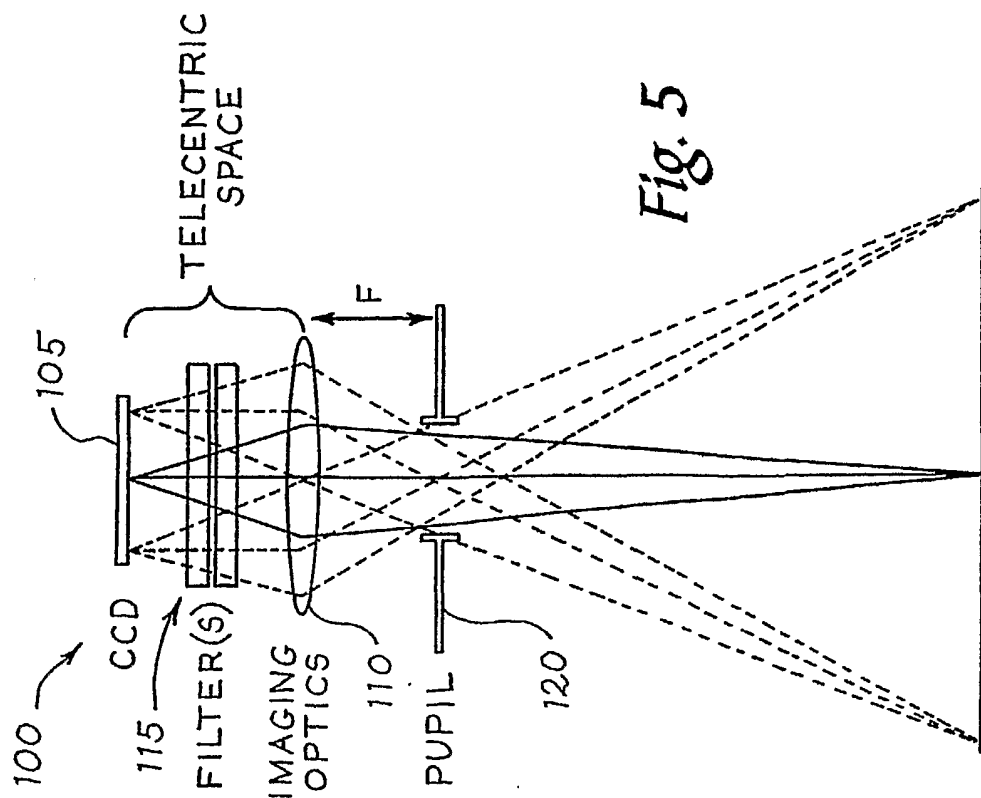


Fig. 1B

--- 0 DEGREES -- 10 DEGREES — 20 DEGREES





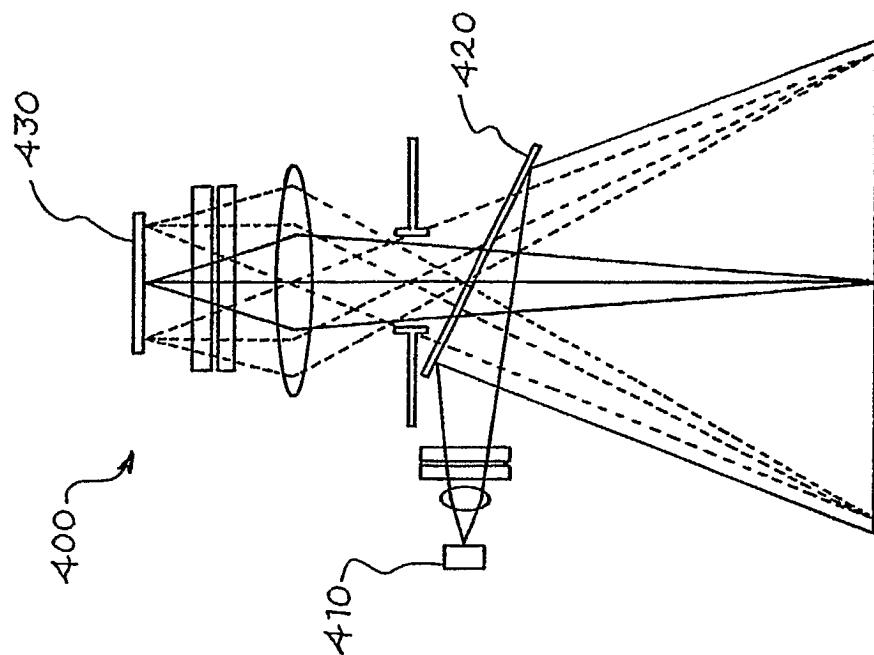


Fig. 8

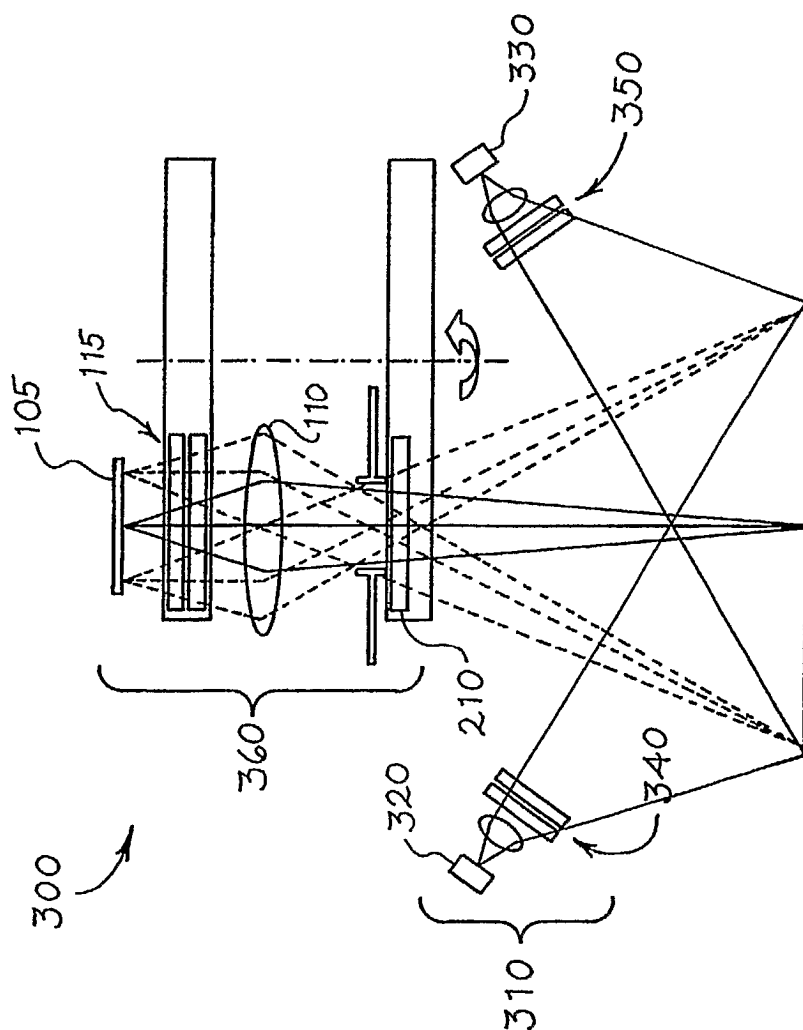


Fig. 7

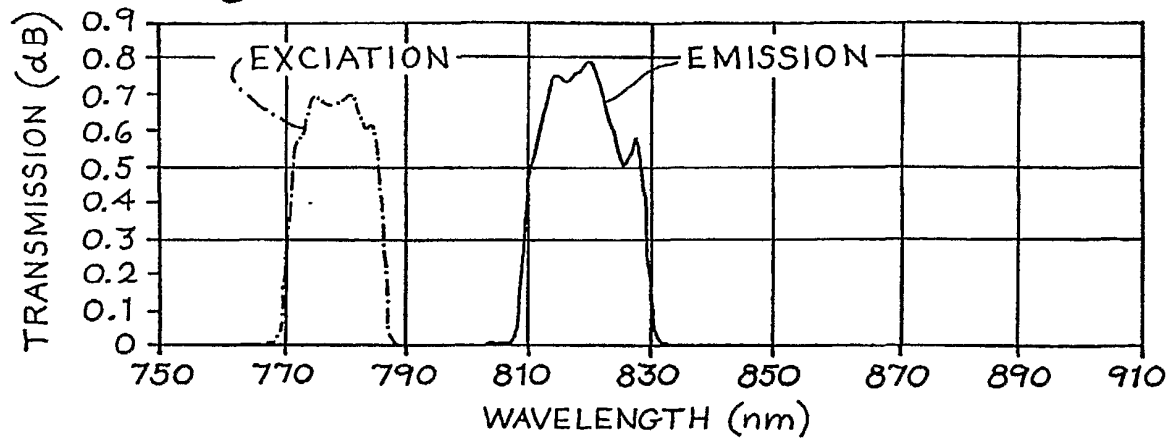
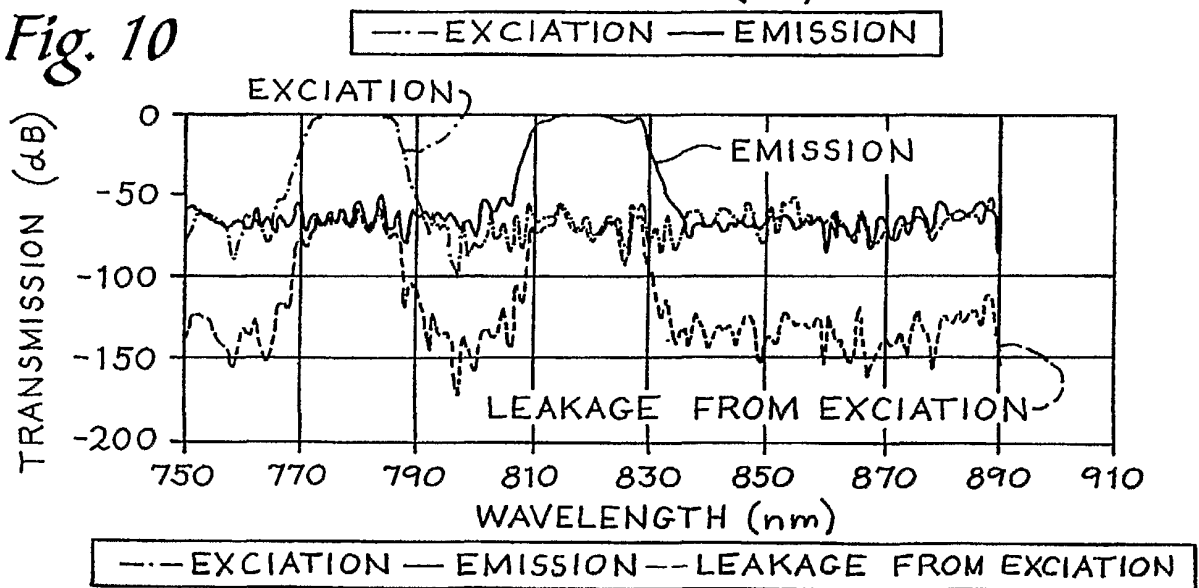
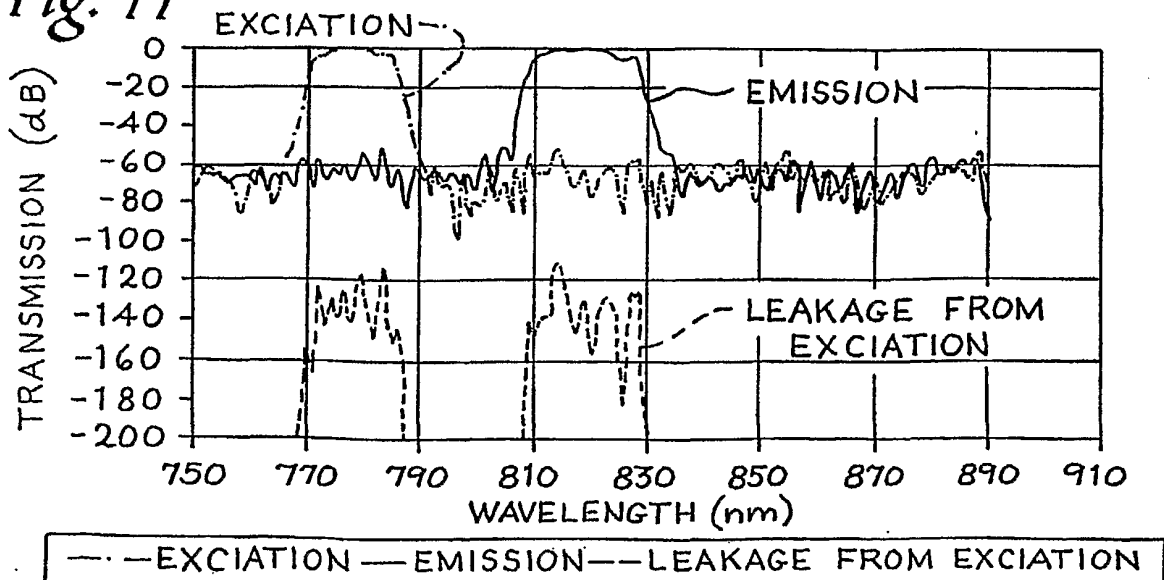
Fig. 9*Fig. 10**Fig. 11*

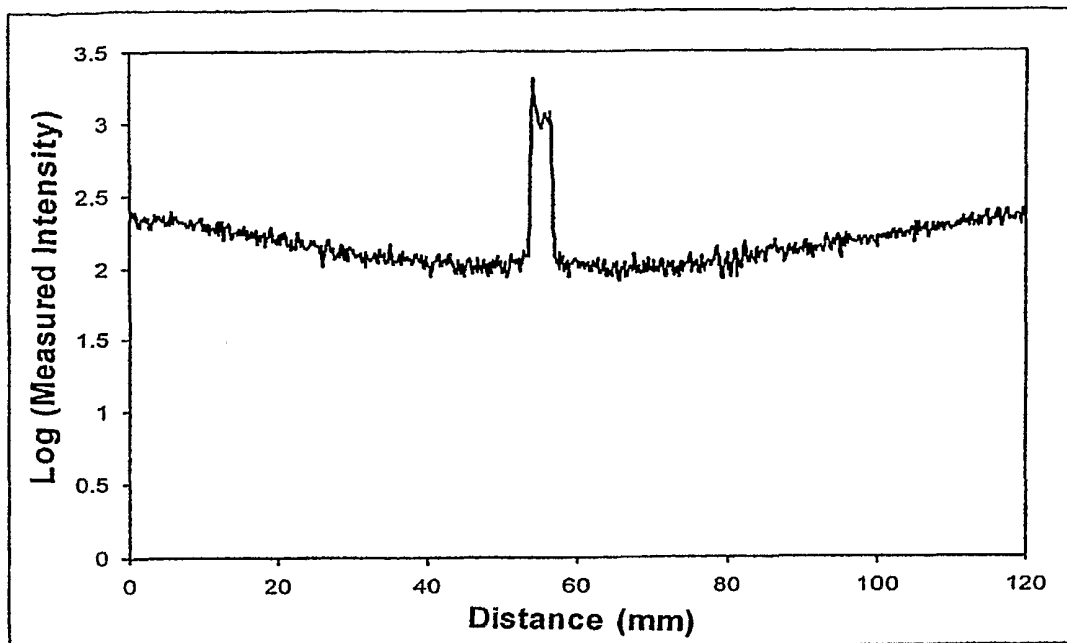
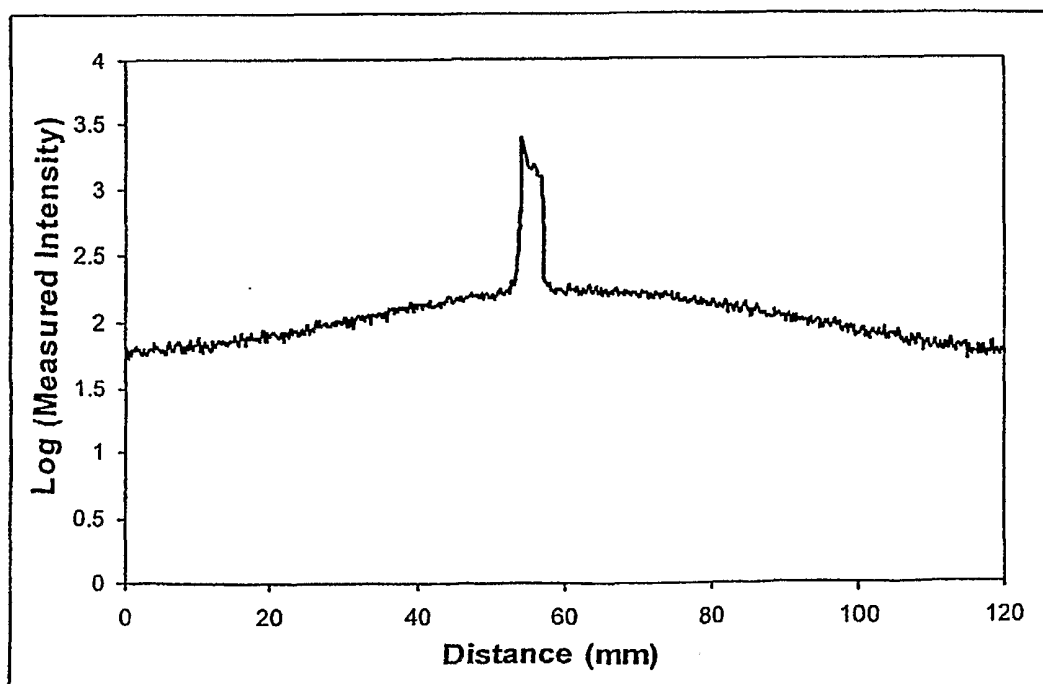
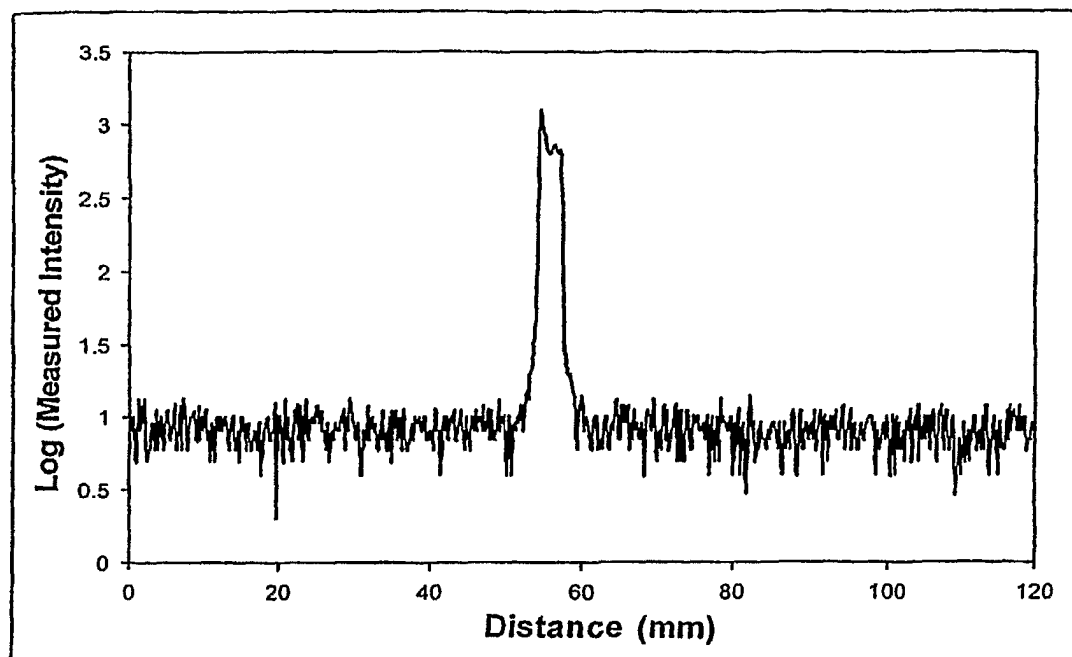
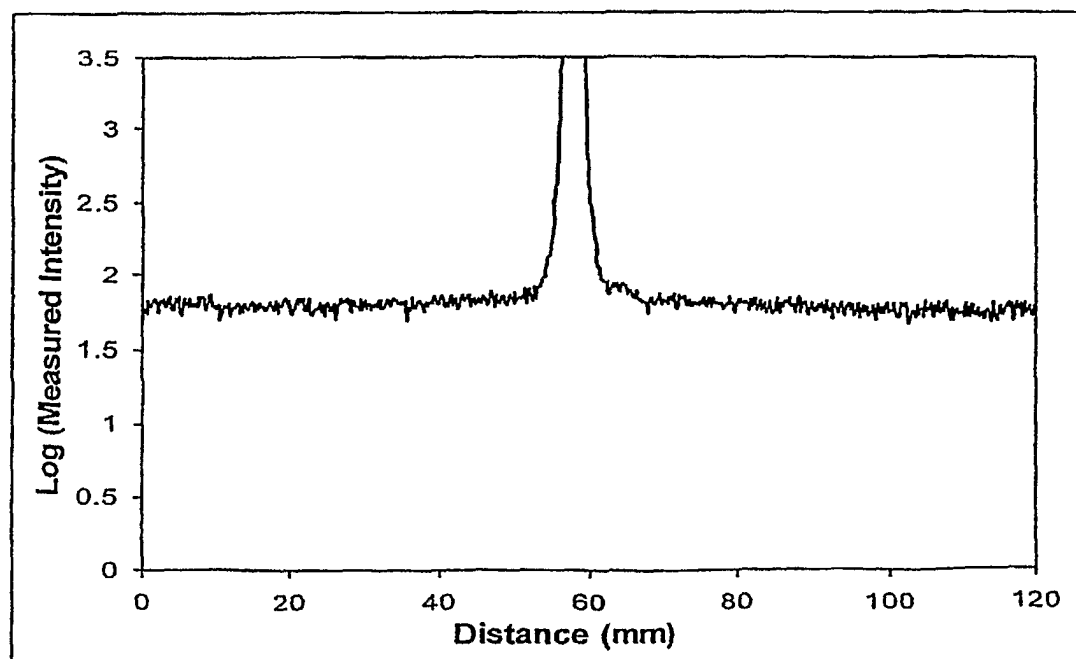
Fig. 12*Fig. 13*

Fig. 14*Fig. 15*

REFERENCES CITED IN THE DESCRIPTION

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